

CLAIMS

1. The use of a *Klebsiella pneumoniae* membrane fraction combined with an antigen or hapten for the preparation of a pharmaceutical composition intended to orient the immune response toward a Th1 type and/or mixed Th1/Th2 type response directed against said antigen or hapten, in which response the Th1 response is close to or greater than the Th2 type response.
2. The use as claimed in claim 1, characterized in that the membrane fraction comprises at least membrane fractions of two different bacterial strains.
3. The use as claimed in either of claims 1 and 2, characterized in that the membrane fraction is prepared by a method comprising the following steps:
- a) culture of said bacteria in a culture medium allowing their growth followed by centrifugation of said culture;
  - b) where appropriate, deactivation of the lytic enzymes of the bacterial pellet obtained in step a), followed by centrifugation of the suspension obtained;
  - c) extraction and removal of nonmembrane proteins and of nucleic acids from the pellet obtained in step a) or b) by at least one cycle of washing the pellet in an extraction solution;
  - d) digestion of the membrane pellet obtained in step c) in the presence of protease enzymes, followed by centrifugation;
  - e) at least one cycle of washing of the pellet obtained in step d) in physiological saline and/or in distilled water; and

- f) ultrasonication of the pellet obtained in step e).
4. The use as claimed in either of claims 1 and 2, characterized in that the membrane fraction is prepared by a method comprising the following steps:
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- a) culture of said bacteria in a culture medium allowing their growth, followed, where appropriate, by centrifugation;
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- b) freezing of the culture medium or of the pellet obtained in step a) followed by thawing and drying of the cells;
- c) removal, by means of a DNase, of the nucleic acids from the dry cells obtained in step b) which have been resuspended;
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- d) grinding of the cells obtained in step c) and clarification of the suspension obtained;
- e) precipitation, in an acid medium, of the suspension obtained in step d) and removal of the pellet;
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- f) neutralization of the supernatant obtained in step e) containing the membrane suspension, followed by dialysis and concentration of the membrane suspension; and
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- g) sterilization of the concentrated membrane suspension obtained in step f).
5. The use as claimed in one of claims 1 to 4, characterized in that said antigen or haptén is chosen from the antigens or haptens specific to an infectious agent or from the antigens associated with tumor cells.
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6. The use as claimed in claim 5, characterized in that said antigen or haptén is chosen from peptides, lipopeptides, polysaccharides, oligosaccharides, nucleic acids, lipids or any
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- 5 compound capable of specifically directing the Th1 type and/or mixed Th1/Th2 type immune response against an antigen or hapten specific to an infectious agent or an antigen associated with a tumor cell.
7. The use as claimed in one of claims 1 to 6, characterized in that said antigen or hapten is coupled or mixed with said membrane fraction.
- 10 8. The use as claimed in one of claims 1 to 7, characterized in that said antigen or hapten is covalently coupled with a supporting peptide to form a complex capable of specifically binding to
- 15 mammalian serum albumin.
9. The use as claimed in claim 8, characterized in that said supporting peptide is a peptide fragment derived from streptococcal G protein.
- 20 10. The use as claimed in either of claims 8 and 9, characterized in that said complex is prepared by genetic recombination.
- 25 11. The use as claimed in one of claims 7 to 10, characterized in that said antigen, hapten or complex is covalently coupled with at least one of the compounds contained in the membrane fraction.
- 30 12. The use as claimed in claim 11, characterized in that the covalent coupling is a coupling carried out by chemical synthesis.
- 35 13. The use as claimed in claim 12, characterized in that there are introduced one or more linking elements into at least one of the compounds contained in the membrane fraction and/or in said

antigen, hapten or complex to facilitate the chemical coupling.

14. The use as claimed in claim 13, characterized in that said linking element introduced is an amino acid.
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15. The use as claimed in claim 11, characterized in that the coupling between said antigen, hapten or complex and at least one of the compounds contained in the membrane fraction is carried out by genetic recombination when said antigen, hapten or complex and said membrane compound are of a peptide nature.
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- 15 16. The use as claimed in one of claims 1 to 15, characterized in that the pharmaceutical composition comprises, in addition, an agent which makes it possible to carry said membrane fraction associated with said antigen, hapten or complex in a form which makes it possible to enhance its stability and/or its immunogenecity.
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17. The use as claimed in claim 16, characterized in that said agent is an oil-in-water or water-in-oil type emulsion.
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18. The use as claimed in claim 16, characterized in that said agent is a particle of the liposome, microsphere or nanosphere type or any type of structure allowing the encapsulation and the presentation in particulate form of said membrane fraction associated with said antigen, hapten or complex.
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- 35 19. The use as claimed in claim 16, characterized in that said agent is chosen from aluminum salts, calcium salts, compounds of plant origin such as

Quil A or saponin, or compounds of bacterial origin such as cholera, pertussis or tetanus toxoid or thermolabile E. coli toxin.

- 5 20. The use as claimed in claims 1 to 19, characterized in that the pharmaceutical composition comprises, in addition, an agent which makes it possible to regulate the immune response induced by said membrane fraction associated with said antigen, hapten or complex.
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21. The use as claimed in claim 20, characterized in that said regulatory agent is chosen from cytokines, growth factors, hormones or cellular components such as nucleic acids, a protein of the family of heat shock proteins or ribosomes.
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22. The use as claimed in one of claims 1 to 21, for the preparation of a pharmaceutical composition intended for the prevention or treatment of infectious diseases or cancers.
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23. The use as claimed in claim 22, characterized in that the infectious disease is of viral, bacterial, fungal or parasitic origin.
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24. The use as claimed in claim 23, for the preparation of a pharmaceutical composition intended for the prevention or treatment of paramyxovirus infections.
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25. The use as claimed in claim 24, characterized in that the paramyxovirus is a respiratory syncytial virus.
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26. The use as claimed in claim 25, characterized in that said antigen associated with the membrane

fraction comprises the peptide G2Na having the sequence SEQ ID No. 4 or one of its homologs whose sequence exhibits a degree of identity of at least 80% with the sequence SEQ ID No. 4.

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27. The use as claimed in claim 26, characterized in that said peptide G2Na or one of its homologs is covalently coupled with a C-terminal fragment (BB) of the streptococcal G protein to form a complex
- 10 capable of binding to mammalian serum albumin.
28. The use as claimed in claim 24, characterized in that the paramyxovirus is a parainfluenzae virus.
- 15 29. A pharmaceutical composition, characterized in that it comprises a membrane fraction prepared by the method as defined in either of claims 3 and 4, and an antigen or hapten associated with said membrane fraction.
- 20 30. The pharmaceutical composition as claimed in claim 29, characterized in that said antigen is chosen from paramyxovirus peptide fragments.
- 25 31. The pharmaceutical composition as claimed in claim 30, characterized in that the paramyxovirus is a respiratory syncytial virus or a parainfluenzae virus.
- 30 32. The pharmaceutical composition as claimed in claim 31, characterized in that said antigen associated with the membrane fraction comprises the peptide G2Na having the sequence SEQ ID No. 4 of the respiratory syncytial virus or a peptide whose
- 35 sequence exhibits a degree of identity of at least 80% with the sequence SEQ ID No. 4.

33. The pharmaceutical composition as claimed in claim 32, characterized in that said peptide G2Na or one of its homologs is covalently coupled with a C-terminal fragment (BB) of the streptococcal G protein to form a complex capable of binding to mammalian serum albumin.
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